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Physicochemical Properties and Interfacial Adaptation of Root Canal Sealers

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This study compared the physicochemical properties and interfacial adaptation to canal walls of Endo-CPM-Sealer, Sealapex and Activ GP with the well-established AH Plus sealer. The following analyses were performed: radiopacity, pH variation and solubility using samples of each material and scanning electron microscopy of root-filled bovine incisors to evaluate the interfacial adaptation. Data were analyzed by the parametric and no-parametric tests (α=0.05). All materials were in accordance with the ANSI/ADA requirements for radiopacity. Endo-CPM-Sealer presented the lowest radiopacity values and AH Plus was the most radiopaque sealer (p=0.0001). Except for ActiV GP, which was acidic, all other sealers had basic chemical nature and released hydroxyl ions. Regarding solubility, all materials met the ANSI/ADA recommendations, with no statistically significant difference between the sealers (p=0.0834). AH Plus presented the best adaptation to canal walls in the middle (p=0.0023) and apical (p=0.0012) thirds, while the sealers Activ GP and Endo-CPM-Sealer had poor adaptation to the canal walls. All sealers, except for ActiV GP, were alkaline and all of them fulfilled the ANSI/ADA requirements for radiopacity and solubility. Regarding the interfacial adaptation, AH Plus was superior to the others considering the adaptation to the bovine root canal walls.

Key Words: physicochemical properties, root canal sealers, scanning electron microscopy, radiopacity, solubility.

Introduction

Root canals sealers can be organized according to their chemical composition. Current materials are based on calcium hydroxide, zinc oxide-eugenol, silicone, polymer resins, glass ionomer (1) and dicalcium silicate, mainly mineral trioxide aggregate (MTA)-derived cements (2). MTA is a widely used biomaterial that includes calcium-based minerals among its main phases. After setting, the material contains calcium oxide that reacts with tissue fluids to form calcium hydroxide (3). However, MTA has not been developed for use as a root canal sealer because of its low flow, reduced workability (short working time and long setting time) and poor consistency (4).

Endo-CPM-Sealer (EGEO SRL, Buenos Aires, Argentina) has been developed to overcome the limitations of MTA, such as poor handling characteristic and lengthy setting time (4), and allow its use as a root canal sealer. This material has similar chemical composition to MTA, but its most significant difference is the presence of a large amount of calcium carbonate, which tends to increase the release of calcium ions, also offering good sealing properties, adhesion to the dentinal canal walls, antimicrobial activity, adequate flow rate and biocompatibility (5-7). The exact proportion of its components is not yet available; however, it is known that it is composed by Portland cement, calcium carbonate, barium sulfate and calcium chloride, to reduce the setting time and improve the handling and sealing properties (6).

The use of glass ionomer sealers is advised based on their adhesion to dentine, fluoride release, biocompatibility and antimicrobial activity. The Activ GP (Brasseler USA, Savannah, GA, USA) filling system consists of glass ionomer-based cement and glass ionomer-coated gutta-percha cones (8). According to the manufacturer, Activ GP has longer handling characteristics, radiopacity, working time and sealing ability compared to previous glass ionomer-based sealers because of its higher flow and slight expansion on setting (9). ActiV GP has been proposed mainly for providing adhesion between the filling material and the root canal walls. It has been evaluated in terms of cytotoxicity (10), microleakage (5,9) and some physicochemical properties. However, the quality of these properties is still in discussion, requiring additional studies in order to complement the available information and to evaluate other not yet studied features.

Sealapex (SybronEndo, Orange, CA, USA) is a calcium hydroxide-based sealer that has good biological properties (11) and apical sealing capacity (5). The manufacturer has recently modified its formulation by adding bismuth trioxide to improve its radiopacity and increase its shelf life, which requires new studies to assess its properties.

AH Plus (De Trey-Dentsply, Konstanz, Germany) is an epoxy resin-based endodontic sealer containing calcium hydroxide, with low solubility and disintegration, adequate radiopacity, adhesion to the root dentine, antimicrobial activity and adequate biological properties (12,13).

One of the factors related to long-term success of endodontic treatment is tridimensional filling and an appropriate coronal restoration, allowing periapical repair and preventing reinfection (14). It is desirable that root canal sealers provide an adherence between gutta-percha (GP) and root canal walls, avoiding the occurrence of gaps at the sealer/dentin interface and providing high level interface adaptability (15). According to Balguerie et al. (16), scanning electron microscopy with longitudinal sections can be used to evaluate the sealer/dentin interface.

The purpose of this laboratory study was to compare the physicochemical properties and interfacial adaptation to canal walls of Endo-CPM-Sealer (based on dicalcium silicate), Sealapex (based on calcium hydroxide - new formulation) and Activ GP Sealer (based on glass-ionomer) with the well-established AH-Plus sealer (based on epoxy resin), using radiopacity, pH and solubility tests according to the American National Standards Institute/American Dental Association - ANSI/ADA - Specification 57 (17) requirements, and scanning electron microscopy (SEM).

Material and Methods

The root canal sealers used in the present study were: Endo-CPM-Sealer (EGEO SRL, Argentina) in powder/liquid ratio 3:1, Activ GP (Brasseler USA, USA) in a 1:3 powder/liquid ratio, Sealapex (SybronEndo, USA) and AH Plus (De Trey-Dentsply, Germany) both in paste/paste ratio. After preparation, the samples were subjected to the analyses described below.

Radiopacity

Polytetrafluoroethylene ring molds (15 mm internal diameter and 1.0 mm high) were used for sample preparation. Five samples per sealer were produced, stored in closed receptacles in an incubator at 37 °C until complete setting. Considering the variations in the setting time of the sealers informed by the manufacturers, the longest setting time of all materials (8 h - AH Plus) was used. Thereafter, the specimens were placed onto 5 occlusal radiographic films (Insight; Kodak Comp., Rochester, NY, USA) alongside a graduated aluminum stepwedge with thickness ranging from 2 to 16 mm, in uniform steps of 2 mm. The x-ray exposures were made using a Spectro II x-ray unit (Dabi Atlante, Ribeirão Preto, SP, Brazil) with a 2.5 mm aluminum filter added. The tube voltage was 70 kV and the current 10 mA. The exposure time was 6.3 s with a constant source-to-film distance of 30 cm. The exposed films were processed manually by the time/temperature method.

The radiographs were digitized using a desktop scanner (Expression 636®; Epson) controlled by software (Epson scanner II 32, version 2.10E®), and then saved in TIFF format. Adobe Photoshop CS3, version 7.0.1. (Adobe System Corporation Inc., San Jose, CA, USA), was used to analyze each image by the intensity histogram of tone scales in the "light channel" to obtain an average value of brightness intensity for each specimen. Contrast and brightness of each image were standardized at 40 and 30, respectively. The radiographic density of the sealers was compared with the radiopacity of different thicknesses of the aluminum stepwedge.

Five repetitions were carried out to determine the radiopacity of the sealers. Data were analyzed statistically by ANOVA and Tukey's test at 5% significance level.

pH

For the pH test, 10 polyethylene tubes measuring 10 mm in diameter and 1.5 mm deep were filled with freshly prepared samples of each material, sealed in flasks containing 10 mL of distilled water, and stored at 37 °C for 30 min. Manual agitation with a glass rod was made to obtain a more homogeneous medium and 2 more minutes were allowed for sedimentation of particles. Next, the pH of solutions was measured with the glass electrode of a digital pH meter (Model DM-20 Digimed; Digicrom Analítica Ltda., São Paulo, SP, Brazil) previously calibrated with buffer solutions with pHs 7.0 and 4.0 at preset times. The pH was measured at the moment of immersion of the material in water and at 1-hour intervals in the first 6 h, and then after 23, 25, 27, 48, 168, 336, 528 and 750 h. The electrode was copiously washed with distilled water and vigorously dried with absorbent paper between readings. The experiment was performed in triplicate and the pH values were recorded for comparison over time.

Solubility

A 1.5-mm-thick cylindrical polytetrafluoroethylene (Teflon; DuPont, HABIA, Knivsta, Sweden) mold with a 7.75 mm inner diameter was filled to a slight excess with freshly mixed sealer (18). The mould was supported by a larger glass plate and covered with a cellophane sheet. A nylon thread was placed inside the material, in order to suspend the samples in water, and another glass plate also covered with cellophane film was positioned on the mould and pressed manually in such a way that the plates touched the entire mould in a uniform manner. The assembly was placed in an incubator (37 °C, 95% RH) for a period 50% longer than the setting time recommended by the manufacturer. As soon as the samples were removed from the mould, they were weighed three times each with 0.0001 g accuracy (HM-200; A&D Engineering, Inc., Bradford, MA, USA) and the mean reading recorded.

The samples were placed in glass flasks containing 7.5 mL of distilled water, taking care to avoid any contact between them and the inner surface of the container, and then kept at 37 °C for 24 h to allow sealer dissolution. After this period, the flasks were centrifuged and the water was poured. After sedimentation of residues of the materials at the bottom of the flasks, they were left at room temperature for several minutes for evaporation of the residual water and then placed in an oven at 110 °C for 30 min to allow complete drying. Then the flasks were reweighed to calculate the sealer mass that was not solubilized in water. The water solubilized mass was calculated based on the initial mass and the final mass. The mass loss of each sample (initial mass minus final mass), expressed as percentage of the original mass was considered as the solubility of the material. The experiment was performed in triplicate. Data were analyzed statistically by the non-parametric Kruskal-Wallis test and Dunn's post-test at 5% significance level.

SEM Analysis of Interfacial Adaptation to Root Canal Dentin

Fifty freshly extracted bovine incisors with fully formed straight roots were disinfected with 2% sodium hypochlorite and stored in saline for 48 h before use. The teeth were randomly assigned to 5 groups (n=10 teeth/group) and crowns were removed at the cementoenamel junction with water-cooled diamond disc at high speed. Each canal was measured by introducing a size 15 K-file (Dentsply-Maillefer, Ballaigues, Switzerland) until its tip was visible at the apical foramen and the working length established at 1 mm short of this point.

Root canal preparation was carried out using a step-back technique. The root canal preparation was performed to the working length up to a size 80 K-file and the root canals were copiously irrigated with 1 mL of 1% sodium hypochlorite at each change of file. After completion of biomechanical preparation, the canals were filled with 1 mL of 17% EDTA for 3 min under agitation, for smear layer removal. Then, the canals received a final flush with 1 mL of 1% sodium hypochlorite, followed by drying with sterile absorbent paper points (Dentsply Ind. and Com. Ltda., Petrópolis, RJ, Brazil). Sealers were mixed for 15-20 s on a clean glass slab using the ratios recommended by the manufacturers.

Obturation techniques with AH Plus, Sealapex and Endo-CPM sealers were performed by introduction into the root canal with a size 80 gutta-percha cone (Tanari Industrial Ltda., Manacapuru, AM, Brazil) up to the working length. Lateral condensation was then completed with the use of finger spreader and accessory points (F and MF gutta-percha points). Regarding the Activ-GP sealer, after placement of the sealer, the Activ GP cone was coated with sealer and slowly inserted into the canal to its working length in order to create a monoblock filling. Radiographs were taken to evaluate the quality of root filling regarding homogeneity and apical extension. Root canal filling was improved if any void was detected radiographically. After filling, the teeth were stored in individual containers and kept at 37 °C (100%) until complete setting, as described before.

Thereafter, the roots were grooved longitudinally with a carborundum disc at low speed and split in the buccolingual plane with a surgical chisel and mallet, taking care not to contaminate the canal with debris. Longitudinal sections were performed according to former studies (16). The buccal and lingual halves were then processed for SEM analysis. The specimens were dried and mounted on aluminum stubs, sputter-coated with a 30-µm-thick gold layer in a fine-coat ion sputter (Denton Desk II, Denton Vacuum LLC, Moorestown, NJ, USA) and examined with a scanning electron microscope (JSM-5410; JEOL Ltd., Tokyo, Japan) operating at 20 kV. The areas of interest in each specimen were selected and images were captured at 15× and 50× magnifications. One calibrated and blinded evaluator examined the SEM micrographs, using a 3-point system to score the interfacial adaptation of the materials to root canal dentin: 0: good adaptation, if well-compacted and tightly adapted filling material was observed, without interface gaps; 1: regular adaptation, if only few interface gaps were found; 2: poor adaptation, if several gaps were found between the sealer and the canal walls or if the filling material was not in contact with the dentin in most parts of the root canal. This analysis was performed after measuring the real root canal length and the value was divided into three thirds. In each third, the median portion was marked to obtain the score.

Data were analyzed statistically by the Kruskal-Wallis test and Dwass, Steel, Critchlow & Fligner post test at 5% significance level.

Results

Radiopacity

The radiopacity values for each root canal sealer are displayed in Figure 1. Endo-CPM-Sealer presented the lowest radiopacity values, followed by Activ GP, Sealapex and AH Plus, which was the most radiopaque sealer. Differences were found between all groups ($p<0.0001$), except between Sealapex and AH Plus ($p=0.5022$).

AH Plus presented the highest radiopacity value and was slightly superior to the last step of the reference stepwedge (16 mm aluminum), while the radiopacity of Endo-CPM-Sealer corresponded to the second step of the scale (4 mm aluminum). According to the ANSI/ADA's Specification Number 57 (17), the radiopacity of a root canal sealer material must be equivalent to at least 3 mm of the aluminum stepwedge.

pH Variation

Figure 2 presents the pH variation of the root canal sealers over time.

AH Plus started with a basic pH (8.82), reached its peak of OH^- ion release after 3 h (mean pH=9.82), and then the pH dropped gradually to 8.57 (23 h) with small variations, remaining around 7.90 until the last reading (31 days) (Fig. 2A).

Sealapex started with a basic pH (7.18), reached its peak of OH^- ion release after 17 h (11.93) and then reduced slightly to around 11.00. From 168 h (7 days), the pH dropped gradually to 8.29 (31 days) (Fig. 2B).

Endo-CPM-Sealer started with a basic pH (9.57) and reached its peak of hydroxyl ion release after 23 h (11.40) and then reduced slightly to around 11.00. From 168 h, the pH dropped gradually to 8.51 (31 days) (Fig. 2C).

Activ-GP was the only material with an acidic pH, starting at 4.48, dropping to 3.83 after 1 h, and remaining without significant changes up to 30 h. Thereafter, the pH increased gradually until reaching a mean value of 5.12 (31 days) (Fig. 2D).

Data interpretation shows that all sealers are basic materials and release OH-ions to the environment, except for ActiV GP, which had an acidic pH from the start of the study (around 4.00). The graphs show a greater release of hydroxyl ions from Sealapex (pH=11.93) followed by AH Plus (pH=9.82) and Endo-CPM-Sealer (pH=11.4), which also presented ion release peaks.

Solubility

Table 1 presents the solubility values (%) for all sealers after the triplicate test. No statistically significant difference (p=0.0834) was found among the sealers. All materials were in accordance with the ANSI/ADA Specification Number 57 (2000) (17) - Guideline for Root Canal Sealers, which requires the solubility to be less than 3 percent in weight.

Interfacial Adaptation to Root Canal Dentin by SEM

Based on the examination of SEM micrographs and statistical analysis of the scores assigned to each material, no statistically significant difference was found among the sealers in the coronal third (p=0.1595), which presented regular adaptation (score 1) to the canal walls. However, in the middle (p=0.0023) and apical (p=0.0012) thirds, there was a statistically significant difference between AH Plus, which had good adaptation (score 0), and sealers Activ GP and Endo-CPM-Sealer, which had poor adaptation (score 2) to the canal walls. Sealapex also presented good adaptation. Figure 3 presents a representative specimen of each root canal sealer.

Discussion

Root filling should ideally have some degree of radiopacity in order to detect its placement (12). In the present work, all sealers fulfilled the ANSI/ADA requirements for radiopacity. Guerreiro-Tanomaru et al. (19) showed that Endo-CPM-Sealer presented radiopacity values above the recommended minimum, equivalent to 6 mm aluminum. However, AH Plus was significantly more radiopaque than

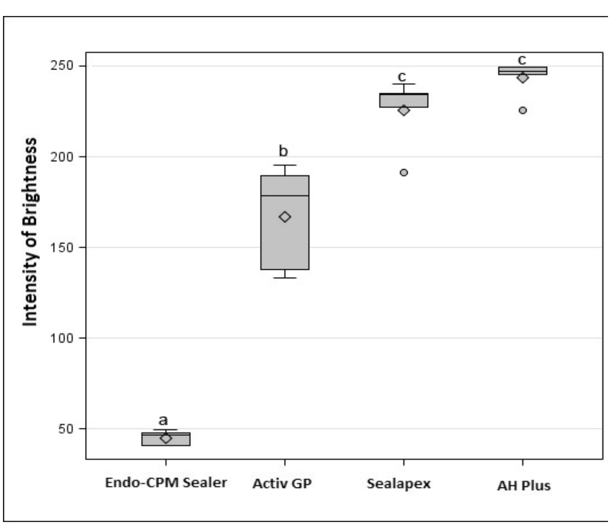

Figure 1. Statistical box plot expressing radiopacity values (intensity of brightness) of Endo-CPM-Sealer, ActiV GP, Sealapex and AH-Plus sealers. Different letters indicate statistically significant difference.

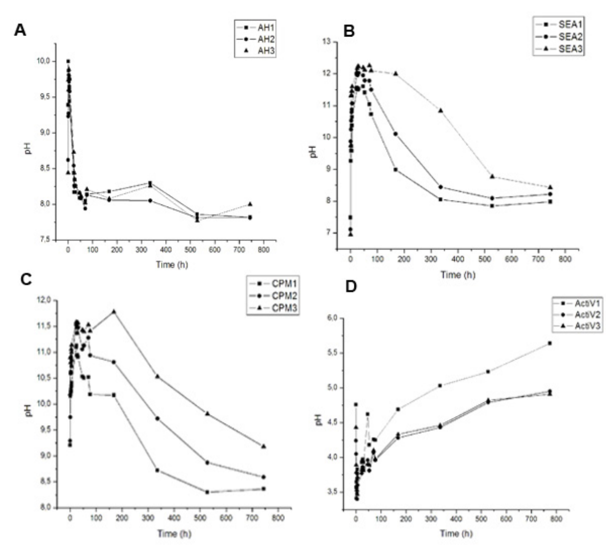

Figure 2. pH variation of sealers AH Plus (A), Sealapex (B), Endo-CPM-Sealer (C) and Activ GP sealer (D) during the experimental period (0 to 750 h).

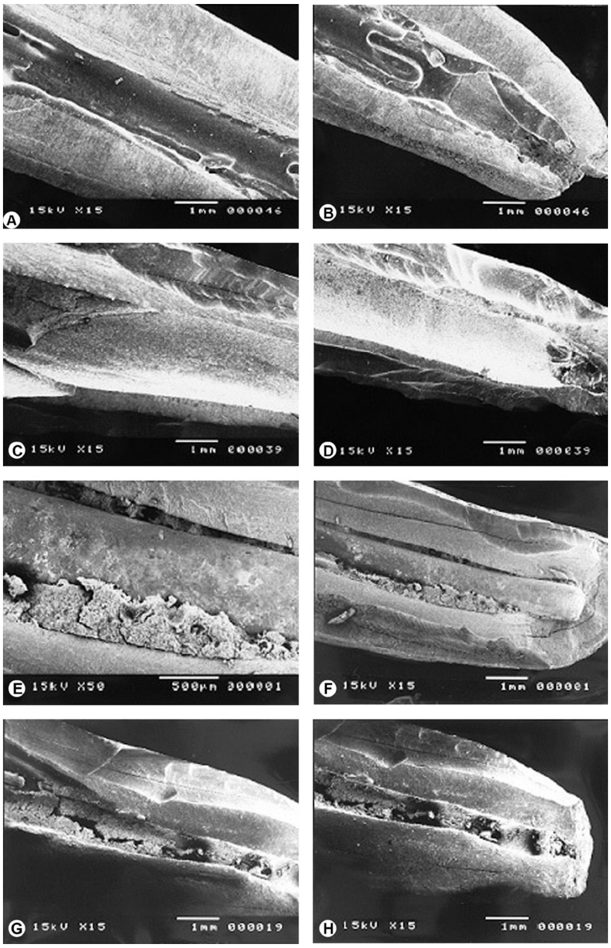

Figure 3. SEM micrographs of representative specimens of AH Plus (A, B), Sealapex (C, D), Activ GP (E, F) e Endo-CPM-Sealer (G, H).

the other materials, which agrees with previous studies that also compared the radiopacity of root canal sealers (12, 20).

In an experiment to compare the radiopacity of various root canal sealers, Tanomaru et al. (21) showed Sealapex to be the least radiopaque, equivalent to 2 mm aluminum, while AH Plus was equivalent to 16 mm aluminum. In the present study, similar results were found for AH Plus (slightly superior to 16 mm), while Sealapex presented significantly higher radiopacity value (14 mm). The difference in the radiopacity of Sealapex is explained by the addition of bismuth trioxide in the recent formulation, which led to a marked improvement in radiopacity. In a more recent study using similar methods and materials, Tanomaru et al. (7) found the radiopacity of Sealapex (new formulation) to be equivalent to 6 mm aluminum, whereas Activ GP sealer glass-ionomer material was equivalent to 2 mm aluminum. Similar result was found by Flores et al. (1), in whose study Activ GP did not fulfill the ANSI/ADA protocols regarding radiopacity. However, in the present study, Activ GP presented high radiopacity, equivalent to 12 mm aluminum.

Periapical healing after root canal therapy should feature bone tissue regeneration, deposition of cementum in the periapex region, and normal periodontal ligament space. Upon contact with water, calcium hydroxide releases calcium ions during ionic dissociation, and the amount of free calcium ions determines the potential of this material to induce formation of mineralized tissue. Moreover, the efficacy of calcium hydroxide is based on its capacity to release hydroxyl ions and cause local increase of pH, which is essential for its clinical antimicrobial action (3).

Root canal filling materials usually contain calcium hydroxide, which is responsible for increasing the pH, although other oxides are also present (3). The composition of these sealers alters the pH according to the reactivity in aqueous medium and/or the pharmacotechnical characteristics of the polymer that composes the material.

Calcium hydroxide may release hydroxyl ions and maintain the pH alkaline and stabilized for a relatively long time. In the present study, the pH measurements revealed that AH Plus, Sealapex and Endo-CPM-Sealer are alkaline materials that

Table 1. Solubility values (%) of the tested sealers.

Sealer	Glass mass	Glass mass + sealer mass (initial mass)	Glass mass + sealer mass (final mass)	Final minus initial mass	Solubility
AH-Plus 1	6.9968	7.0497	7.0008	0.0489	0.69%
AH-Plus 2	6.9238	7.1039	7.0371	0.0668	0.94%
AH-Plus 3	6.9248	7.1810	7.1808	0.0002	0.002%
Sealapex 1	6.9011	7.0321	7.0288	0.0033	0.04%
Sealapex 2	6.8921	7.0216	7.0128	0.0088	0.12%
Sealapex 3	6.8141	7.1162	7.1218	0.0056	0.07%
ActiV GP 1	6.9308	7.0252	6.9941	0.0311	0.44%
ActiV GP 2	6.9084	7.0622	7.0218	0.0404	0.57%
ActiV GP 3	8.4347	8.7374	8.6100	0.1274	1.46%
Endo-CPM Sealer 1	6.9210	7.0305	6.9770	0.0535	0.76%
Endo-CPM Sealer 2	8.1998	8.3230	8.2531	0.0699	0.84%
Endo-CPM Sealer 3	6.7989	6.9651	6.8922	0.0729	1.05%

release hydroxyl ions to the medium, which is in accordance with previous studies (11). However, what is seen on Figures 2A-2C is a rapid increase in pH values followed by a decrease tending to stabilization. This may be explained by the fact that calcium hydroxide solubility decreases as the sealer sets and the already released hydroxyl ions are consumed by other substances present in the solution or on sealer surface.

ActiV GP was the only sealer that was acidic from the start, with pH values close to 4.0. This was an expected result because this material derives from glass ionomer cement, which is composed by polyacrylic and itaconic acids that release H^+ ions, increasing the acidity. The analysis of pH variation of this sealer (Fig. 2D) shows an initial decrease followed by increase, indicating that as observed for alkaline sealers, the H^+ ions are consumed by other components in the solution.

Solubility is the capacity of a substance to dissolve into another, and is expressed as the concentration of the saturated solution of the former in the latter (22). All sealers tested in this experiment were in accordance with the ANSI/ADA requirements, as they presented solubility less than 3%. Sealapex presented mean solubility of only 0.055% while AH Plus was the most soluble (0.82% mean value). These results do not agree with those of a recent study (2) in which Sealapex was shown to have solubility values above the limit established by ANSI/ADA, whereas AH Plus was in accordance with this ANSI/ADA requirement for solubility. According to Eldeniz et al. (11), Sealapex was also considered to present high solubility, however this statement was assigned due to its high pH and high calcium ion release. However, the low solubility of Sealapex in the present study is justified by its composition, which was formulated with the purpose of providing greater radiopacity and low solubility, without losing the good biological properties.

Correlation of the solubility profile with pH variation over time revealed that the most soluble sealer, AH Plus, also had a faster consumption of hydroxyl ions (Fig. 2A). Therefore, it could be inferred that there is a secondary reaction between the release of hydroxyl ions and the subsequent reaction with the sealer byproducts dissolved in aqueous medium.

High solubility is beneficial both from physicochemical and biologic viewpoint, because the release of more calcium ions into the tissue, as well as the higher pH may lead this sealer to exhibit a more powerful antibacterial effect (10).

In spite of having low solubility and diffusibility, calcium hydroxide will not promote the expected healing effects on surrounding tissues, unless calcium and hydroxyl ions dissociate from the sealer. This is why the long-term sealing ability of calcium hydroxide sealers and their therapeutic effects are subject to great discussion (3).

Good adaptation to root dentin is an important property of endodontic sealers for providing a tight seal and preventing bacterial leakage and failure of treatment (23). In the present study, in the middle and apical thirds, AH Plus and Sealapex had good adaptation, while Activ GP and Endo-CPM-Sealer presented poor adaptation to the canal walls. The result of the present study can be correlated with the findings of Balguerie et al. (16), who showed that AH Plus presented optimal tubular penetration and adaptation to the root canal wall, after SEM evaluation. There are no studies in the literature regarding root canal adaptation after SEM evaluation of Activ GP, Endo-CPM and Selapex sealers.

It is noteworthy that bovine teeth were selected for this study, as they are readily available and share basic microscope morphologic qualities (24), being a good substitute for human dentin. Also, according to Schilke et al. (25), there are no significant differences in dentinal tubule diameters in human and bovine dentin observed by SEM.

In conclusion, all sealers met the ANSI/ADA requirements for low solubility and radiopacity greater than 3 mm of equivalent aluminum. Except for the glass-ionomer sealer (Activ GP), all materials were neutral and became basic when dispersed in water, although the effect reduced over time. Additionally, in SEM examination, the AH Plus material was superior to the others for adaptation to the bovine root canal walls.

Resumo

Este estudo comparou as propriedades físico-químicas e a adaptação interfacial às paredes do canal dos cimentos Endo-CPM-Sealer, Sealapex e Activ GP com o bem estabelecido cimento AH Plus. As seguintes análises foram realizadas: radiopacidade, variação de pH e de solubilidade utilizando amostras de cada material, e microscopia eletrônica de varredura utilizando incisivos bovinos obturados para avaliar a adaptação interfacial. Os dados foram analisados utilizando testes paramétricos e não-paramétricos (α=0,05). Todos os materiais estavam de acordo com os requerimentos da ANSI/ADA para radiopacidade, sendo que o Endo-CPM-Sealer apresentou os menores valores de radiopacidade e o AH Plus foi o cimento mais radiopaco (p=0,0001). Exceto o Activ GP, que foi ácido, todos os outros cimentos apresentaram natureza química básica e liberaram íons hidroxila. Com relação à solubilidade, todos os materiais estavam de acordo com as recomendações da ANSI /ADA, sem diferença significante entre os cimentos (p=0,0834). O AH Plus apresentou a melhor adaptação às paredes do canal nos terços médio (p=0,0023) e apical (p=0,0012), enquanto que os cimentos Activ GP e Endo-CPM-Sealer apresentaram uma pobre adaptação às paredes do canal. Em conclusão, todos os cimentos, exceto o Activ GP, foram alcalinos e todos preencheram os requerimentos da ANSI/ADA para radiopacidade e solubilidade. Com relação à adaptação interfacial, o AH Plus foi superior aos demais para adaptação às paredes do canal radicular de incisivos bovinos.

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